ijms-390001

Supplementary data

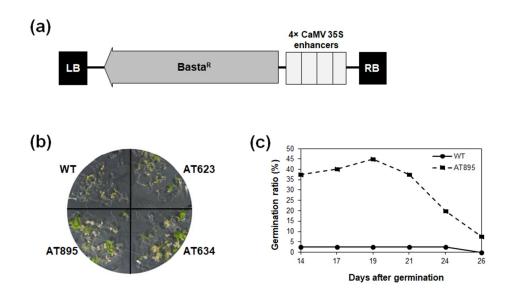


Figure S1. Isolation of AT895 from activation tagging lines. (a) Schematic map of the activation tagging vector, pFGL942. Four copies of *CaMV 35S* enhancers were used for the activation of genes. Basta-resistant gene was used as a selective marker for transgenic plants. (b) Germination of WT and T₂ plants of AT895, AT623, and AT634 on 210 mM NaCl-containing MS agar media. (c) Germination ratio of WT and T₂ plants of AT895 on 210 mM NaCl-containing MS agar media up to 26 DAG.

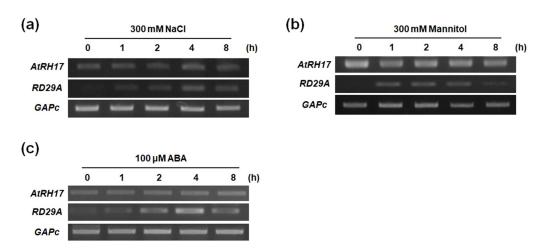


Figure S2. Expression analysis of AtRH17 under osmotic stresses using semi-quantitative RT-PCR. (a) Expression of AtRH17 under 300 mM NaCl treatment for 0, 1, 2, 4, and 8 hr. (b) Expression of AtRH17 under 300 mM mannitol treatment for 0, 1, 2, 4, and 8 h. (c) Expression of AtRH17 under 100 μ M ABA treatment for 0, 1, 2, 4, and 8 hr. GAPc was used as an internal control. At least two biological replicates showed similar results, with one shown here.

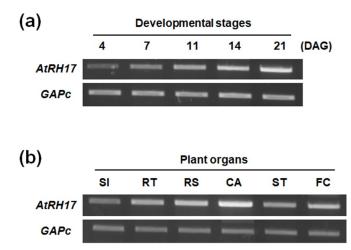


Figure S3. Analysis of temporal and spatial expression patterns of *AtRH17* using semi-quantitative RT-PCR. (a) Semi-quantitative RT-PCR analysis of *AtRH17* in 4-, 7-, 11-, 14-, and 21-day-old WT seedlings grown under SD conditions. *GAPc* was used as an internal control. (b) Semi-quantitative RT-PCR analysis of *AtRH17* expression in organs of 36-day-old WT grown under LD conditions. *GAPc* was used as an internal control. SI, siliques; RT, roots; RS, rosette leaves; CA, cauline leaves; ST, stems; FC, floral clusters. At least two biological replicates showed similar results, with one shown here.

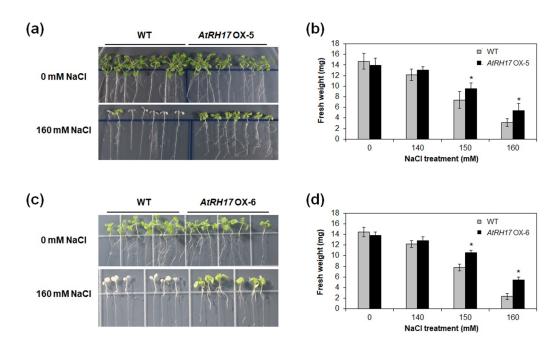


Figure S4. Salt-stress response of AtRH17 OX seedlings. (**a**,**c**) Responses of WT and AtRH17 OX T₃ seedlings to 0, 140, 150, and 160 mM NaCl. Five-day-old seedlings were transferred onto NaCl-containing MS agar media and photographs were taken 10 days after NaCl treatments. (**b**,**d**) FW was measured 10 days after NaCl treatments. Error bars represent the standard deviation (n = 35 plants) and * indicate t-test P < 0.05.

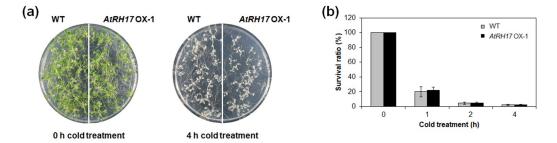


Figure S5. Cold-stress response of AtRH17 OX seedlings. (a) Responses of WT and AtRH17 OX-1 T₃ seedlings to freezing treatment for 0 and 4 hr. Three-week-old seedlings on MS agar media were kept at -8 °C, and photographs were taken after five days of recovery at 22 °C. (b) Survival ratio was measured after five days of recovery. Error bars represent standard deviation (n = 25 plants). Three independent T₁ lines showed similar results, with one shown here.

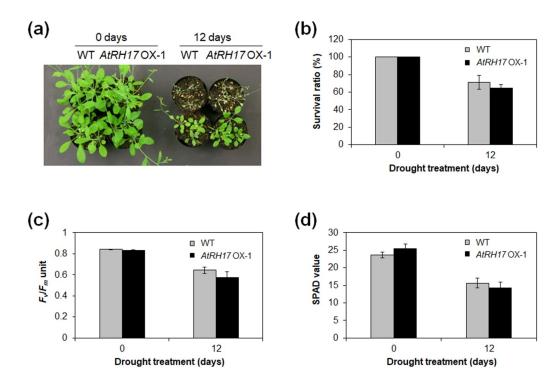


Figure S6. Drought-stress response of AtRH17 OX mature plants. (a) Three-week-old WT and AtRH17 OX-1 plants were dried for 12 days and then rewatered. Photograph was taken after five days of rewatering. (b) Survival ratio of WT and AtRH17 OX-1 dried for 12 days and rewatered for five days. (c) PS II activity (F_v/F_m) of WT and AtRH17 OX-1 dried for 12 days and rewatered for five days. (d) SPAD values of WT and AtRH17 OX-1 dried for 12 days and rewatered for five days. Error bars represent standard deviation (n = 30 plants). Three independent T₁ lines showed similar results, with one shown here.

Table S1. List of primers used for PCR.

Gene	Forward	Reverse	Purpose
AtRH17	5'-TTCCCATACCCGGCAATATG-3'	5'-TCGGCGTAAGTAAGGGATTC -3'	Quantitative RT-PCR
GAPc	5'-GTGTCCCAACCGTTGATGTC-3'	5'-TCCCTTGAGTTTGCCTTCGG-3'	Quantitative RT-PCR
RD29A	5'-CCTGAAGTGATCGATGCACC-3'	5'-CAGTGGGTTTGGTGTAATCG-3'	Quantitative RT-PCR
RAB18	5'-TACCAGAACCGTCCAGGAGG-3'	5'-CGTACTCGTCATACTGCTGC-3'	Quantitative RT-PCR
RD29B	5'-TTCTTGGCTCGGTGGTAAAC-3'	5'-GGTGCCAAGTGATTGTGGAG-3'	Quantitative RT-PCR
RD22	5'-GTAAACCCGGTAAAAGAACC-3'	5'-TACACGAAAGGGTTTGCTCC-3'	Quantitative RT-PCR
COR47	5'-ATGTACCAGTTTCCACTACC-3'	5'-TCCTCTGCTTTCTCGTCGTG-3'	Quantitative RT-PCR
DREB2A	5'-GTGTTGCCAACGGTTCATAC-3'	5'-GAGGTATTCCGTAGTTGAGG-3'	Quantitative RT-PCR
DREB2B	5'-GAAGAGTCTTGTGGAACCAG-3'	5'-CCCAATACTGCTGCTCAAAC-3'	Quantitative RT-PCR
AtRH17	5'-TTCTGAGACAGAAGAGGAGG-3'	5'-TCGGCGTAAGTAAGGGATTC-3'	Semi-quantitative RT- PCR
GAPc	5'-CACTTGAAGGGTGGTGCCAAG-3'	5'-CCTGTTGTCGCCAACGAAGTC-3'	Semi-quantitative RT- PCR
RD29A	5'-GAAACAGAGTCTGCCGTGAC-3'	5'-TGCTGCCTTCTCGGTAGAGA-3'	Semi-quantitative RT- PCR
AtRH17 OX	5'-GTG <u>GTCGAC</u> ATGAAG AGAGCCCAACAATC-3'	5'-CGC <u>GGATCC</u> AGTTTT TTGTGTACTTCTAT-3'	Cloning
sGFP- AtRH17	5'-GTG <u>GTCGAC</u> ATGAAG AGAGCCCAACAATC-3'	5'-CGC <u>GGATCC</u> GAGTTT TTTGTGTACTTCTAT-3'	Cloning
AtRH17- sGFP	5'-GTG <u>GTCGAC</u> ATGAAG AGAGCCCAACAATC-3'	5'-CGC <u>GGATCC</u> TCAAGT TTTTTGTGTACTTC-3'	Cloning